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PURPOSE: Immortalized cell lines representing fibroblast cells from AB corneal stroma would facilitate studies of corneal cell biology and injury response. METHODS: Primary cultures of cells derived from mouse corneal stroma were transfected with a human telomerase reverse transcriptase (hTERT) expression construct to maximize chances of cellular immortalization. A resulting cell line was analyzed for telomerase activity, cell growth characteristics, senescence and gene expression patterns. Specific responses to transforming growth factor beta (TGF-beta) were also analyzed. RESULTS: An immortalized cell line was derived and was named MK/T-1. MK/T-1 cells show no signs of cellular senescence or transformation at over 100 passages. Telomerase activity was significantly higher in MK/T-1 cells as compared to the parental cell cultures. However, relative telomere length (RTL) in the MK/T-1 and parental cells was not significantly different. Senescence associated beta-galactosidase (SA-beta-Gal) activity was not detected in late passage MK/T-1 cells while the parental cells had already upregulated SA-beta-Gal at high levels by passage 9. The MK/T-1 cells express vimentin, tubulin, lumican, mimecan, decorin and collagen I, but not keratocan. Exposure of the MK/T-1 cells to TGF-beta induces the expression of smooth muscle alpha-actin (ASMA), the activation of MAP Kinase (p38-MAPK) and morphological changes consistent with cytoskeletal reorganization. CONCLUSIONS: MK/T-1 cells represent an immortalized fibroblast cell line derived using cultures from corneal stroma cell preparations. Expression of hTERT

using cultures from corneal stroma cell preparations. Expression of hTERT may contribute to immortalization of the MK/T-1 cells by a mechanism other than increases in RTL. MK/T-1 cells may be a useful model in which to study the responses of corneal fibroblast cells to cytokines and other diverse environmental factors in vitro.

L7 ANSWER 2 OF 26 MEDLINE

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TITLE: Excitability and contractility of skeletal muscle

engineered from primary cultures and

cell lines.

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AB The purpose of this study was to compare the excitability and

contractility of three-dimensional skeletal muscle constructs, termed myooids, engineered from C2Cl2 myoblast and 10Tl/2 fibroblast cell lines, primary muscle cultures from adult C3H mice, and neonatal and adult Sprague-Dawley rats. Myooids were 12 mm long, with